

Unknown lab



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The possible identity of the unknown organisms in the mixed culture was limited to bacteria that we had worked with previously in lab. Initially a Gram stain was conducted in order to distinguish the unknown bacterium as a Gram-positive and/or a Gram-negative organism (Lancaster and Bennett, 2012; Kellenberger, 2001). Based upon the results, both Gram-negative and Gram-positive bacteria were observed in the unknown mixed culture.

In order to isolate the two different bacteria, colonies that grew on the MSA were used to inoculate Gram-positive tests, where as MacConkey Agar colonies were used to inoculate Gram-negative tests. Once the colonies were isolated and the appropriate Gram-negative and Gram-positive tests were conducted, the identification of both unknown organism were fairly easy. The results from the Coagulase test, the Oxidase test, and the Nitrate test were used to help identify the unknown Gram-positive bacterium.

The coagulase test was negative, therefore of all the possible Gram-positive bacterium the unknown organism could either be *Staphylococcus simulans* or *Micrococcus luteus*. Next, the Nitrate test was used to differentiate between the two remaining possibilities. Since the unknown Gram-positive bacterium resulted in a negative for Nitrate, the organism was identified as *Staphylococcus simulans*.

As for the identification of the unknown Gram-negative bacterium, the unknown bacterium's ability to ferment lactose was analyzed first. Positive lactose fermentation was indicated on both the MacConkey agar plate and SSA by the growth of bright pink colored colonies. Identical results were also found in the inoculated lactose broth with the Durham tube and the TSI tests, which both indicated positive fermentation for lactose as well.

According to the results from the data collected, the unknown Gram-negative bacterium was identified as a lactose fermenter and therefore the unknown bacterium could be any of the Enterobacteriaceae including *Escherichia coli*, *Enterobacter aerogenes*, *Klebsiella pneumoniae*. To differentiate between these three Enterobacteriaceae and identify the #8 unknown organism IMViC tests results were analyzed next (2012; Kellenberger, 2001). Positive methyl red results indicated the unknown's ability to produce acids when degrading glucose, causing the pH to drop. Of the possible Enterobacteriaceae, *Escherichia coli* are the only bacterium that produces acids (Watson, 2013). Comparatively, *Enterobacter aerogenes* and *Klebsiella pneumoniae* produce more neutral products from glucose and therefore test negative for methyl red (Anonymous, 2013e). Therefore according to the results collected the unknown Gram-negative bacterium was identified as *Escherichia coli*.

Additionally, to further support our identification, negative results from the citrate test indicated the unknown organism's inability to utilize citrate. These findings supported the elimination of *Enterobacter aerogenes* and *Klebsiella pneumoniae* which both test positive, being able to utilize citrate as its sole carbon and energy source. Also, indole positive results indicated the unknown bacterium's production of tryptophanase and therefore indole. Indole production also supports the identification of the unknown Gram-negative as *Escherichia coli*.

Escherichia coli are part of the Enterobacteriaceae family, Gram-negative bacteria that grow in the intestinal tract of humans and other animals. *Escherichia coli* is the only coliform that is found only in feces, and is not naturally environmental, as the other coliforms occasionally are.

Microorganisms such as bacteria are responsible for many diseases, water contamination, and spoiling of food, although they are also very beneficial while sharing symbiotic relationships with humans and other organisms (Watson, 2013).

Identification of bacteria is of great importance in many fields such as health care and environmental microbiology. There are many reasons behind the importance of knowing the identity of microorganisms including the identification of the causative agent of an illness in a patient. For example being able to identify *Klebsiella pneumoniae* as the cause of a patient's illness can lead to appropriate and rapid treatment for that specific organism.

Additionally, the identification of specific bacteria and their characteristics can also be useful in knowing the correct microorganism to be used for making certain antibiotics. This lab was helpful in the identification of an unknown bacterium. Tests conducted were important in the microbiology lab because they allow researchers to identify the organisms they are working with. By being able to identify the bacterium, researchers can identify other characteristics of that specific organism, in addition to a proper diagnosis and treatment.